pending upon whether the infection is local or systemic. Local tuberculosis such as lupus, glands, early tuberculosis of the lungs, early tuberculosis of the bone, show a low index. The late tuberculosis of the lung shows a variable index, sometimes as high as two or three times the normal; at others equally low. The practical point in all this work lies in the relation of the opsonins to treatment. Wright and Ross and Bullock, treating large numbers of cases of different kinds of tubercular infections by inoculation methods controlled by opsonin determinations, have had much success. The same is true of Hollister in this country.

The idea of treatment is to increase the opsonins in the patient's blood so that the bacteria meeting these will become an easy prey to the white cells which have heretofore not been able properly to perform their work of phagocytosis. The method has been especially successful in cases of lupus. The plan is as follows:

The patient's opsonic index is determined: he is then given a measured amount of a vaccine composed of the dead and sterilized bodies of tubercle bacillus. These dead cultures are readily obtainable in the market in the form of Koch's tuberculin R. You will say that Koch's tuberculin antedates the opsonic theory and vaccinations with tuberculin R., have often proven disastrous to patients; this is very true, but any very powerful drug used empirically may become destructive to patients. The great value of Wright's method is that he is enabled to measure the effects of the dose and to give the tuberculin in a rational as opposed to an empirical manner. Having determined the opsonic index of his patient, he inoculates not to exceed 1-1000 of a milligram of dry tuberculin and then he takes the opsonic index of that patient to the tubercle bacillus daily.

It is the invariable result of the first inoculation that the opsonic content drops very markedly and remains low for some time, varying with the different type of case and with the individual. The drop in the opsonic content Wright calls the negative phase, but this within a short time rises and goes well above the normal. Now if while the patient's opsonins are rapidly descending, a second injection is given there will be a still further decrease in the opsonic power and a further injection still may overwhelm the patient and make him unable to fight the invading process. But if the opsonic index is watched and the second inoculation is given while the opsonic index is rising above normal, there will be a still greater rise and the defensive power will become much increased and the patient as a rule will go on to a better condition. The value of the opsonic method is here evident as a control to the inoculation.

From the work that is being done we are coming to learn that not only tubercular and staphylococci infections may be combatted, but a case Ross has recorded of a chronic empyema due to pneumococcus which cleared up promptly under vaccinations with dead pneumococci, controlled by opsonic determinations to pneumococci, shows that this method will be valuable in localized pneumo-infections. Other work done leads us to hope that the chronic joint infections due to invasions by Neiser's coccus may also soon be efficiently treated.

In fact, there is every reason to hope that in vaccination controlled by opsonin determinations we have a therapeutic method that will give us power against many of the infections over which we now have no control. Besides the therapeutic gain, the diagnostic value is great as evidenced by a case of sycosis Wright reports in which, when he found the opsonic index high for staphylococcus, he tried it for tubercle and, finding that index low, was enabled to diagnose a tubercular sycosis and to cure it with tuberculin. Ross' case of pneumococcus infection is another, and I have no doubt cases of hip joint disease will be earlier and more positively diagnosed because of our ability to determine this index. It has already been shown that children of tubercular mothers have on the average an index but half that of those whose mothers are not infected, and that the index of bottle-fed infants against a number of bacteria is less than that of the breast-fed. And so we have explained rationally a well-known clinical fact. Undoubtedly we will soon test out the effect of many remedies on the activating power of the serum. It will be interesting to know whether the coloid metals gain their potency as agents in septicemia by a power to increase the opsonic activity of the serum. So high an authority as Welch has proclaimed Wright's discovery the greatest in medicine since Koch gave us knowledge of the tubercle bacilli and the means to isolate and study them. Personally I think that he understates the importance of the work, and that we have in the method of Wright and Douglas a technic that will overturn much of our therapy and supplant it with a method of treatment at once rational and efficient.

OPSONIC TECHNIC.*

By LEWIS SAYNE MACE, M. D., San Francisco.

The method of estimating the opsonic content of blood serum elaborated by A. E. Wright of London is briefly as follows:

The washed white blood cells, or leukocyte cream, is prepared by drawing ten or fifteen drops of blood from a normal individual into a small centrifuge tube filled with a 1.5 per cent. sodium citrate solution in .85 per cent. sodium chloride, shaking thoroughly and centrifuging about five minutes. The citrate solution is pipetted off and .85 per cent. sodium cholride added, the tube thoroughly shaken and again centrifuged. The red cells, being heavier, are thrown to the bottom of the tube and on top of these the leukocytes are seen as a thin gray film. These washed corpuscles are pipetted off and placed in a watch glass.

The bacterial emulsion in the case of tubercle bacilli is made by removing the growth from a culture with a platinum loup and thoroughly grinding

* Read at the Polyclinic Gathering.

up in a morter moistened with 0.1 per cent. sodium chloride solution, drops of the sodium chloride solution are added from time to time and the grinding continued until the clumps of bacilli are broken up as thoroughly as possible. The emulsion is then placed in a test tube and heated to 100 degrees for five minutes and sealed until ready for use.

When preparing for an estimation, the tube containing the emulsion is thoroughly shaken and centrifuged a few minutes to thrown down the clumps. The upper layer, which should be quite opalescent, is pipetted off and placed in a watch glass.

The serum to be tested is obtained by making a light stab with a sharp capillary glass tube in the top of a finger near the nail. The blood which flows readily, is collected in a glass capsule having curved capillary ends, which are sealed when the capsule is about two-thirds filled with blood. It is then allowed to coagulate and hung by its curved arm over the ring of the centrifuge and revolved until the clear serum has collected above the clot. The top is now broken off, the serum removed with a capillary pipette and placed in a third watch glass properly marked. For the control test a glass capsule is filled in the same way with blood of several normal individuals and centrifuged as before. The blood of a number of normal people is used as a control to correct the error of individual variation in opsonic content.

The three preparations, washed white corpuscles, bacterial emulsion and serum, are measured and mixed with a capillary pipette having a mark about two centimeters from the end. The blood cells are drawn up to the mark, then a bubble of air is allowed to enter the tube and the same volume of bacterial emulsion drawn in, then another space of air is allowed to enter the tube and an equal volume of the serum to be tested drawn up and the three volumes carefully mixed by drawing up and down several times upon a slide. Finally the mixture is drawn into a pipette, the end of which is sealed in the flame and placed in the incubator with the control tube, which is prepared in the same way at the same time. After incubating fifteen or twenty minutes, the tube end is broken off and the mixture again mixed by drawing up and down on a slide. Two slides are prepared by smearing as in preparing blood smears, except that the preparation is made much thicker and allowed to dry slowly in the air. In this way the leukocytes can easily be found collected at the outer edge of the smear and counted. The slides are fixed in saturated bichloride of mercury for one minute and stained in carbofuchsine, destained in dilute sulphuric acid, and counterstained in methylene blue.

In counting, it is necessary to count only polynuclear leukocytes and to count only those in which the neucleus lies flat. In this way the error of counting is reduced to a minimum.

The best emulsion of tubercle bacilli is on which in the ordinary time of incubation shows one or two bacilli per cell and which does not contain clumps of bacilli. The length of time necessary for grinding and centrifuging each culture of tubercle bacilli used must be ascertained by practice tests before beginning estimation.

DISCUSSION OF PAPERS BY DRS. PORTER AND MACE.

Dr. Evans—I think we ought to congratulate Dr. Mace on the degree to which he has perfected this technic. No one knows, except those who have tried it, the discouragements met with in this work. There are few points in the technic where he differs from Wright. For instance, the last part of his demonstartion regarding the collection of the blood. I should imagine that he would be very apt to burn the blood in this way. Dr. Wright on the contrary, after allowing the blood to run into the capsule, heats the empty portion of it before sealing that end. Then the area cooling the blood is drawn from the capsular end. This work is very fascinating, and anyone who has had the privilege of seeing Wright at work in his laboratory is impressed with the seriousness of the man and with the fact that his demonstrations have been very valuable contributions to modern scientific medicine. It would be impossible for such a one to fail to be convinced that he has demonstrated the existence of these bacterio-tropical substances in the blood serum. It is particularly on the lines of tuberculosis that this work has assumed importance, bringing forth as it does acute indications for exact dosage of tuberculin or other culture products used therapeutically. By following the opsonic curve of individuals under treatment, Wright has demonstrated the fact that much smaller doses of tuberculin are indicated than had been used by those who have relied on clinical evidence alone. A good deal of the work done in London both by Wright and Bullock, who is bacteriologist at the London Hospital, and is an ardent supporter of this work, has been done in the treatment of lupus. Their results with tuberculin in this disease have convinced them that treatment with Finsen light, X-ray and other irritating conditions are of value only by reason of the fact that they produce a hyperemia of the affected part, thereby flooding the lesion with blood rich in opsonic content. I recall one case of a girl with lupus, under Dr. Bullock's treatment, who had a very persistent lesion in spite of her opsonic index having been raised above normal as the result of the use of tuberculin. An ordinary old-fashioned poultice was sufficient to flood this lesion with opsonines, thereby causing very rapid recovery, when the fact was appreciated that hyperemia of the lesion was all that was necessary.

The practical difficulty in the way of the use of this valuable discovery in private practice is the difficulty, the result of the complexity of technic, and it is to be regretted that until this technic is very much simplified the opsonic work of Wright can not be placed in daily use by the busy clinician.

Dr. Porter—When you have a patient in whom you suspect tuberculosis, a low or variable opsonic index will give you confirmatory evidence. Wright has especially called attention to the fact that at times when a number of joints are affected, gentle massage of a single joint will cause not only that joint but other affected joints to improve. He attributes this to the fact that the massage has inoculated the patient to a slight degree. It is of some interest to know that it has been shown recently by Ross that an infant born of a tubercular mother has an opsonic index about equal to the mother's, which gives us an explanation why tuberculosis has been considered hereditary.

Amburg has shown that a bottle-fed baby has an opsonic index to all infections lower by far than a breast-fed baby. These facts explain a great deal